Granulocyte Colony-stimulating Factor Producing Anaplastic Carcinoma of the Pancreas: Case Report and Review of the Literature

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Abstract. We report on the case of a 67-year-old man with granulocyte colony-stimulating factor (G-CSF) producing anaplastic carcinoma of the pancreas. Preoperative routine tests revealed an elevated white blood cell (WBC) count of 25.2 G/l, consisting almost exclusively of neutrophilic granulocytes (23.31 G/l) with a predominance of segmented neutrophils (78% of all neutrophilic granulocytes), and elevated levels of C-reactive protein at 87 mg/l. Upon surgery, local tumour infiltration was more extensive than expected from preoperative imaging. However, no peritoneal dissemination was found and curative resection was attempted. Only seven days after the operation, signs of relapse were seen upon computed tomography. Histology revealed an undifferentiated anaplastic carcinoma, on the basis of a poorly differentiated ductal adenocarcinoma. Immunohistochemistry demonstrated G-CSF and G-CSF-Receptor expression in some CD68-positive syncytial macrophages. Granulocyte colony-stimulating factor (G-CSF) in serum was elevated at 5.6 pg/ml, which further raised to 43 pg/ml one week after FOLFIRINOX chemotherapy (oxaliplatin, irinotecan, 5-fluorouracil), while WBC decreased from 103.3 G/l to 59.3 G/l. Granulocyte macrophage-colony stimulating factor (GM-CSF) in serum was normal (<0.5 pg/ml). The patient died on postoperative day 34.

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G-CSF producing pancreatic cancer is a rare tumour entity with dismal prognosis. No standard treatment has been established so far. Further studies are needed to elucidate the potential autocrine stimulation through co-expression of G-CSF and G-CSF-Receptor and the different cellular sources and role of G-CSF in the aggressive nature of the disease. Neutrophilia, due to the production of cytokines such as G-CSF and GM-CSF in solid tumours, is a rare albeit well-described phenomenon. To our knowledge, only 13 cases have been reported so far, including our case.

Case Report

We report on a case of a 67-year-old man who was diagnosed with anaplastic carcinoma of the pancreatic tail. The patient reported a four-month history of left upper abdominal pain, loss of appetite, and weight loss of 9 kg. Computed tomography (CT) showed a 7-cm lesion in the pancreatic tail and enlarged peripancreatic lymph nodes but no signs of distant metastasis (Figure 1). The patient was planned to undergo resection with curative intent. Preoperative routine tests revealed an elevated white blood cell count (WBC) of 25.2 G/l (Figure 2) and an elevated C-reactive protein (CRP) of 87 mg/L (data not shown). Leukocytosis consisted almost exclusively of neutrophilic granulocytes (23.31 G/l) with a predominance of segmented neutrophils (78% of all neutrophilic granulocytes, data not shown).

Upon laparotomy, no peritoneal dissemination was found. However, local tumour infiltration was more extensive than expected based on preoperative imaging. Multivisceral resection was performed including left pancreatectomy, splenectomy, partial gastrectomy and resection of the left colonic flexure. Despite a favourable postoperative course, the patient's WBC continued to increase after a nadir of 24 G/l on the fourth postoperative day (Figure 2). Suspecting bacterial infection, empirical antibiotic treatment with

Figure 1. Computed tomography of a 67-year-old man diagnosed with anaplastic carcinoma of the pancreatic tail. A 7-cm lesion in the pancreatic tail (white arrows) and enlarged peripancreatic lymph nodes (black arrows) can be seen.

piperacillin-tazobactam was started and switched to meropenem-vancomycin when WBC further increased. However, the patient was in good condition and had no fever. Repeat blood cultures did not show growth of any germs. CT scan was repeated seven days after the operation, showing new enlarged lymph nodes with central necrosis retropancreatically and thus confirming fast growth of relapsed tumour.

Histology revealed an undifferentiated anaplastic carcinoma, on the basis of a poorly differentiated ductal adenocarcinoma (maximum diameter 13.5 cm) with diffuse infiltration of the peripancreatic tissue, lymphovascular and perineural infiltration as well as breaching of the serosa. Union for International Cancer Control (UICC) TNM-classification was pT3 pN1 (24/35) pM1 (5/5 peritoneal lymph nodes). Tumour grade was G4 and surgical margins were not clear microscopically (R1). Immunohistochemical staining analysis of serial sections was performed (Figure 3). Giant cells were seen upon haematoxylin-eosin (HE) staining (Figure 3A) and since they were CD68-positive might represent syncytial macrophages (Figure 3B), some of them staining positive for granulocyte colony-stimulating factor (G-CSF, Figure 3C), while approximately half of them staining positive for granulocyte colony-stimulating factor-receptor (G-CSF-R, Figure 3D). G-CSF was also elevated in serum at 5.6 pg/ml (Figure 2), confirming the paraneoplastic nature of neutrophilia with ectopic production of G-CSF. GM-CSF in serum was normal (<0.5 pg/ml). Therefore, antibiotics were stopped. The patient recovered well from the

Figure 2. White blood cell counting (WBC, $\times 1000$ cells/ μ l=G/l; left vertical axis) and granulocyte colony-stimulating factor levels (G-CSF, pg/ml; right vertical axis) in the serum before and after the operation as well as at the first cycle of FOLFIRINOX chemotherapy. Even though WBC count decreased after both, the operation and the chemotherapy, an increase of G-CSF was observed after the 1st cycle of chemotherapy.

operation despite further increase in WBC count and was discharged on the 19th postoperative day in good clinical condition. Total WBC count at the time of discharge was 76 G/l (Figure 2).

The patient was started on chemotherapy 24 days after the operation. The FOLFIRINOX regimen was chosen as it was shown to be associated with a survival advantage compared to gemcitabine single agent chemotherapy (1). Treatment consisted of oxaliplatin 85 mg/m² IV on d1, irinotecan 180 mg/m² IV on d1, and 5-fluorouracil IV bolus 400 mg/m² on d1 and as a continuous infusion of 1,200 mg/m²/d on days 1 and 2, repeated every two weeks (1). Prior his 1st cycle of chemotherapy, the patient's WBC increased to 103.3 G/l. Chemotherapy was well tolerated without significant side effects during the first week. On day 8 of chemotherapy, WBC decreased to 59.3 G/l but, interestingly, serum G-CSF increased to 43 pg/ml (Figure 2). Unfortunately, the patient decided to stop chemotherapy for personal reasons, was lost to follow-up and died shortly thereafter, on postoperative day 34.

Discussion

G-CSF induced neutrophilia is commonly seen in carcinomas of the lung and in undifferentiated carcinoma. Patients with paraneoplastic leukaemoid reactions typically are clinically stable despite having large tumour burdens. However, clinical outcomes are poor unless effective antineoplastic treatment is received (2). To our knowledge, only 13 cases

Table I. Reported cases of granulocyte colony-stimulating factor producing pancreatic cancer.

Case	Age (yrs)	Gender	Symptoms	WBC (G/L)	Neutrophilic granulocytes (%)	G-CSF (pg/mL) (normal value)	Cellular localisation of G-CSF	CRP (mg/dl)	Histology	Treatment	Survival (days)
(17)	71	m	ND	71.3	ND	650	ND	ND	Pancreatic carcinoma	ND	ND
(16)	72	f	ND	61	91	86 (<60)	ND	ND	ND	ND	ND
(18)	64	m	Fever	24.3	93	157 (<30)	(Negative for all cells)	0.62	Anaplastic carcinoma	Operation	56
(12)	83	m	Back pain	15.7	81	123 (6.0-21.9)	Squamous carcinoma cells (but not adenomatous)	ND	Poorly differentiated adenosquamous carcinoma	Chemotherapy: 5-fluorouracil, pirarubicin, mitomycin-C	120
(14)	60	m	Weight loss, fever	28.9	70	77 (<30)	Adenocarcinoma cells	15.16	Poorly differentiated adenocarcinoma	Palliative care	46
(7)	50	m	Leukocytosis	49.18	ND	350 (5-15)	Adenocarcinoma cells	ND	Poorly differentiated adenocarcinoma	Palliative care	20
(15)	46	m	General fatigue	14.3	83.7%	155 (5.78-27.5)	ND	4.6	Anaplastic carcinoma	Operation	120
(9)	56	m	Back pain, fever	18.7	81	62	Tumour cells	13.99	Pancreatic ductal adenocarcinoma	Chemotherapy: S-1 gemcitabine	135
(13)	74	m	Fever, general fatigue	29.5	ND	110 (<18.1)	Pancreatic cancer cells (cytoplasmic)	15.9	Poorly differentiated adenocarcinoma	Operation	42
(11)	63	m	Weight loss, upper abdominal pain	91.5	87.5	134 (<18.1)	Pancreatic cancer cells	ND	Anaplastic pleomorphic-type carcinoma	-	11
(8)	89	f	Weight loss, fever	45.86	96	690 (<18.1)	Atypical cells (adenosquamous carcinoma)	11.98	Poorly differentiated adenosquamous carcinoma	Palliative care	62
(10)	68	m	Fever, weight loss	17.5	ND	355 (<39)	Tumour cells	13.2	Anaplastic carcinoma	Chemotherapy: S-1	83
Our case	67	m	Weight loss, upper abdominal pain	25.2	92.5	5.6	Syncytial macrophages	87	Undifferentiated anaplastic carcinoma	Operation and chemotherapy: oxaliplatin, irinotecan, 5-fluorouracil	34

of G-CSF producing pancreatic cancers have been reported summarized in Table I.

In these published cases, mean age of patients was 66 years (range, 46 to 89 years), with a predominance of male vs. female patients (85% vs. 15%). Most patient presented with fever, back and/or upper abdominal pain and a history of weight loss. WBC count at the time of diagnosis ranged from 14.3 to 91.5 g/l, while 70 to 91% of the white blood cells consisted of neutrophilic granulocytes. In most patients, mild elevations of CRP were seen, which were not as prominent as one would expect in a bacterial infection with similar high WBC count. Histologically, most patients presented with anaplastic or poorly differentiated adenocarcinoma, while two patients suffered from poorly differentiated adenosquamous carcinoma. The clinically aggressive course of disease is in line with the poorly differentiated/anaplastic histology of all tumours, if reported, and is reflected by the dismal prognosis. In 4 of 13 patients an operation could be attempted, and only 4 of 13 patients

received chemotherapy. Median survival of the published cases of G-CSF producing pancreatic cancer was 56 days, which is considerably shorter than the 2 to 6 months seen in stage IV pancreatic cancer in general (3).

In our patient, an attempt was made using a relatively aggressive triplet of chemotherapy (oxaliplatin, irinotecan, and 5-fluorouracil), resulting in a reduction of WBC from 103 to 59 G/l within one week. However, since the patient decided to stop all therapy despite good tolerance to the chemotherapy, it's not possible to know if this more aggressive chemotherapy regimen would have resulted in better efficacy than single agent chemotherapy (1). Two observations are of interest in our case: the increase of serum G-CSF levels after chemotherapy, while at the same time WBC count decreased, and immunohistochemical localisation of both, G-CSF and G-CSF-Receptor in syncytial macrophages.

An inverse relationship between endogenous G-CSF serum levels and circulating neutrophil counts has been reported, which might explain the increase of G-CSF one

Figure 3. Histochemistry and immunohistochemistry of serial sections (100 $\times$). (A) Giant cells in an undifferentiated anaplastic carcinoma of the pancreas are seen upon haematoxylin-eosin (HE) staining. (B) Macrophages and giant cells stained positive for CD68. Giant cells, thus, are syncytial macrophages. (C) Some of the CD68-positive giant cells stain positive for G-CSF. (D) Approximately half of the CD68 positive giant cells stain positive for G-CSF-Receptor.

week after chemotherapy in our patient, despite a decreased in WBCs (4, 5). Since the first description of plasma G-CSF elevation in nude mice transplanted with human lung cancer (6), more than 100 cases of G-CSF producing tumours have been described, including virtually all solid tumours and some haematological neoplasias. While cells of the monocyte/macrophage lineage are among the most prominent sources of G-CSF, malignant cell lines derived from numerous cell types have been shown to be capable of constitutively secreting large amounts of G-CSF (4). In 8 of the 12 published cases of G-CSF producing pancreatic cancer, G-CSF could immunohistochemically confined to tumour cells (7-14). In three, no staining results were reported (15-17) and in one, staining remained negative for G-CSF in all cells (18). However, the ability to produce G-CSF is characteristic of a variety of cell types following

appropriate stimulation, including tumour necrosis factor (TNF) (19), interleukin (IL) 1 (20, 21), IL3 (22), IL4 (23), GM-CSF (22, 24) and interferon- γ (IFNG) (25). It is possible that in our patient's tumour, some of the syncytial macrophages were stimulated to produce G-CSF. The expression of both, G-CSF and G-CSF-Receptor by the same cells in G-CSF producing tumours has only been described once, in a case of hepatocellular carcinoma (26).

The poor prognosis of G-CSF producing pancreatic cancer may be linked to the elevated G-CSF itself. In fact, a correlation was found between breast cancer cell-derived G-CSF, the production of atypical T-cell-suppressive neutrophils (27), and the overexpression of G-CSF augmenting tumour growth (28), further corroborating the well-described relationship between an increase of circulating neutrophils and poor prognosis in cancer

patients (29). In summary, we report on a patient with G-CSF producing pancreatic cancer, a rare tumour entity with dismal prognosis. No standard treatment has been established so far. Further studies are needed to elucidate the potential autocrine stimulation through co-expression of G-CSF and G-CSF-Receptor and the different cellular sources and role of G-CSF in the aggressive nature of the disease.

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